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Fundamental Investigation of Laccase Mediator Delignification
on High Lignin Content Kraft Pulps

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FUNDAMENTAL INVESTIGATION OF LACCASE MEDIATOR DELIGNIFICATION ON HIGH LIGNIN CONTENT KRAFT PULPS

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ABSTRACT

The fundamental bleaching chemistry of high and low-kappa softwood (SW) kraft pulps via laccase-mediator systems (LMS) was explored. The high-kappa pulps responded more favorably to LMS than the low-kappa pulps. The residual lignin from the low and high kappa pulps was enriched in acid groups after LMS(E*) treatments. Accompanying this effect was a decrease in the phenoxy content. This decrease was more pronounced in the case of the low kappa pulps.

INTRODUCTION

The bleaching of kraft pulps is an integral component in the production of high-value paper products. Over the last decade, research efforts have been largely focused on environmental issues, but as these challenges have been addressed, new research opportunities are being developed. Kraft pulping and bleaching yields have historically been important research topics that have now taken on renewed interest. Improved pulp yield offers the opportunity to enhance kraft wood utilization practices and reduce operating and capital costs associated with the production of kraft pulps. Pulp yields can be improved by a variety of techniques including modified chipping techniques, pulping additives, and halting the kraft pulping process at high kappa numbers. The latter process attempts to utilize the high selectivity of the initial and bulk phase of a kraft cook. Typically, the cook is halted at or before reaching the residual phase. This yields SW kraft pulps with kappa numbers in the range of 40-50. The remaining lignin is then removed by oxygen delignification. Jameel et al. (1) and others (2, 3) have shown that this approach can effectively improve the overall yield of bleached kraft pulp by 2-3 %.

Recently, our research group has begun to explore several competing technologies that

could increase pulp yields. One attractive approach is to use biobleaching technologies as a potential cost-effective alternative to oxygen delignification. The high selectivity of LMS delignification of low kappa kraft pulps (kappa # > 30 for SW and > 15 for HW) has been established by several researchers (4,5,6,7). In addition, the LMS stage has been shown to remove more than 45% of the residual lignin present in low-lignin kraft pulps. By extension, we had hypothesized that an LMS stage could remove significant amounts of lignin from high-kappa kraft pulps with high selectivity. The results of our initial investigations supported our hypothesis that an LMS stage could be employed to remove lignin from high-kappa kraft pulps (8). Figure 1 summarizes some of the results of applying an LMS stage to a SW kraft pulp with an initial kappa of 97.5.

This report further examined the chemistry involved in delignification of high-kappa kraft pulps. A series of LMS-stages were performed on a 75.4-kappa conventional southern SW kraft pulp and a 33.8-kappa conventional southern SW kraft pulp. Both pulps originated from the same tree. The pulps were then extracted using an E, EO, EP, and EPO stage. The nature of the residual lignin was also examined.

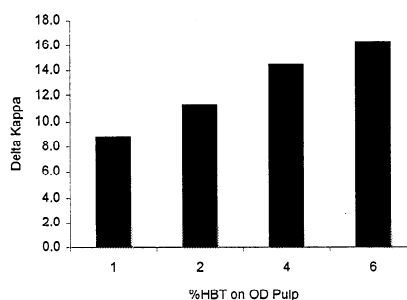


Figure 1. Effect of HBT charge on delignification of a 97.5 kappa SW kraft pulp with constant laccase charge.

EXPERIMENTAL

Materials

All materials were purchased from Aldrich and used as received except for *p*-dioxane, NHAA, and laccase. *p*-Dioxane was freshly distilled over NaBH₄ prior to using it. NHAA was synthesized in accordance with Oxley's method (9). Laccase, from *Trametes villosa* was donated by Novo Nordisk Biochem.

Enzyme Assay

Laccase activity was measured by monitoring the rate of oxidation of syringaldazine.

One unit of activity (U) was defined as the change in absorbance at 530 nm of 0.001 per minute, per ml of enzyme solution, in a 100 mM phosphate buffer (2.2ml) and 0.216 mM syringaldazine in methanol (0.3ml). The procedure was carried out at 23°C.

Laccase-Mediator Delignification Procedure

A 2000 mL capacity Parr reactor, equipped with a pressure gauge, a stirrer, a heating jacket, and a digital thermometer, was charged with 60 g of never-dried pulp (solid basis). The pulp consistency was adjusted to 9% by adding distilled water. The slurry was then heated to a temperature of 45°C and was maintained at this temperature for the duration of the enzymatic treatment. A 2% dose of HBT (on o.d. weight) was then added (when NHAA was used, a 2% dose was added on the same molar equivalence as HBT) to the heated slurry. Subsequent to mixing the slurry (about 5 minutes), the pH was adjusted to 4.5 with glacial acetic acid. Laccase was then added (9.2 mg of laccase per 10 g of o.d. pulp). The reactor was then sealed and pressurized with oxygen to 145 psig. Subsequent to the four hour treatment, the pulp was thoroughly washed with water and subjected to various reinforced alkali stages (E*). All extraction stages were conducted at 10% consistency for one hour and at 80°C. The E* stages are summarized in Table 1. Kappa and brightness measurements were performed on the extracted pulps in accordance with T236 and T212 (10), respectively.

Table 1 Summary of E*-conditions

| Extraction stage | % NaOH (o.d. basis) | % H ₂ O ₂ (o.d. basis) | O ₂ (psig) |
|------------------|---------------------|--|-----------------------|
| E | 2.5 | - | - |
| EO | 2.5 | - | 60 |
| EP | 2.5 | 0.5 | - |
| EPO | 2.5 | 0.5 | 60 |

Isolation of Residual Lignins

The isolation of residual lignin from kraft pulps was carried out in accordance with standard literature methods (11). A 5L three-necked round bottom flask equipped with a Friedrichs condenser was charged with 50 g of o.d. pulp (air-dried). The consistency of the pulp was adjusted to 4% by adding a 0.10N(HCl) 9:1 *p*-dioxane: water solution. The slurry was then refluxed for 2 hours under an argon atmosphere. Subsequent to the treatment, the pulp was filtered and the filtrate was filtered through celite. The filtrate was then neutralized and concentrated under reduced

pressure to approximately 10% of the original volume. Water (400 ml) was added and the mixture was concentrated again under reduced pressure. The solution's pH was then adjusted to 2.5 with 1.00N HCl. The precipitate (i.e., the lignin) was collected, washed several times, and freeze-dried.

Characterization of Residual Lignins

The residual lignins isolated from the untreated brownstock (33.8 kappa and 75.4 kappa) and from LMS_{HBT}(E), LMS_{HBT}(EPO), LMS_{NHAA}(E), and LMS_{NHAA}(EPO) treated pulps were characterized by ³¹P NMR. The NMR data was acquired with a DMX400 MHz Bruker spectrometer. The ³¹P NMR experiments were conducted in accordance with standard literature methods (12).

RESULTS AND DISCUSSION

Based on our previous studies, we were confident that an LMS stage could be used to delignify high-kappa kraft pulps. As the next step in investigating LMS delignification of high lignin content kraft pulps, we proceeded to examine the chemistry of LMS delignification of low (kappa # 33.8) and high (kappa # 75.4) lignin content kraft pulps. Although a variety of compounds can be employed in an LMS stage, two of the most effective mediators we have are 1-hydroxybenzotriazole (HBT) and N-acetyl-N-phenylhydroxylamine (NHAA). The low-and high-kappa kraft pulps were delignified under exactly the same LMS conditions using NHAA and HBT. Subsequent to the LMS stage, the pulps were extracted with an E, EO, EP, and EPO-stage. The changes in delignification and brightness gains are summarized in Figures 2-5.

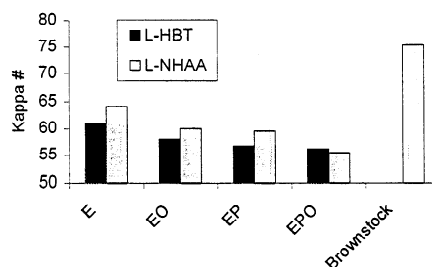


Figure 2: Delignification of the kappa 75.4 SW kraft pulp with LMS followed by E* stage.

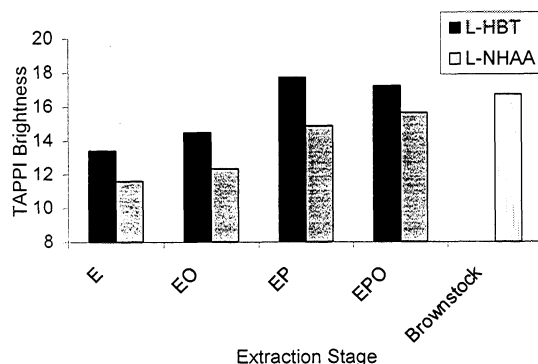


Figure 3: TAPPI brightness of the kappa 75.4 SW kraft pulp treated to LMS(E*).

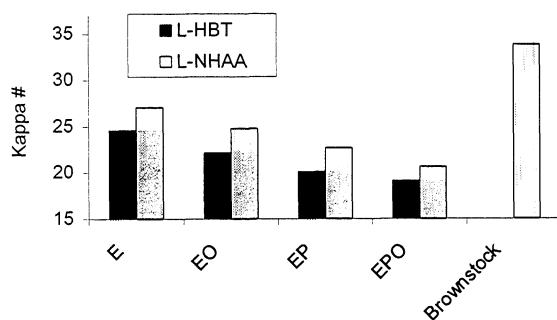


Figure 4: Delignification of the kappa 33.8 SW kraft pulp with LMS followed by E* stage.

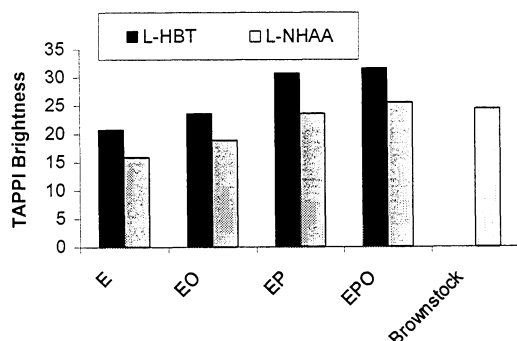


Figure 5: TAPPI brightness of the kappa 33.8 SW kraft pulp treated to LMS(E*).

The results from LMS delignification of the high- and low-kappa pulps bear a strong resemblance. In each case, the use of HBT as a mediator provided superior delignification under our experimental conditions. The use of an oxidatively reinforced alkaline extraction stage improved the observed delignification effects. Interestingly, the use of hydrogen peroxide in an EP-stage was found to substantially improve the delignification effect. This differs from the response frequently observed with D_0 pulps in which the EP-stage principally brightens the pulps. It is also interesting to note that the use of an EPO stage narrows the difference of the kappa number for the HBT and NHAA LMS-treated pulps. Nonetheless, the high-kappa pulp always exhibited an improved bleachability after LMS(E*) in comparison to the low-kappa pulp.

Treatment of the high and low kappa pulps with either HBT or NHAA in an LMS stage results in a darkening of the kraft pulps. Pulp brightness values did not respond favorably to either an E or EO, stage but the use of peroxide in an EP and EPO yielded a favorable response in brightness. Clearly, the use of peroxide in an EP or EPO stage results in lignin and chromophore removal.

Residual Lignin Analysis

To further explore the fundamental principles involved in controlling the observed delignification effects for the LMS(E*) stages reported in Figures 2–5, we elected to isolate lignin from the starting low- and high-kappa brownstocks. In addition, the residual lignin from the LMS(E) and LMS(EPO) stages was isolated. In all cases, the lignin samples were isolated from the pulp using an acidic *p*-dioxane/water extraction procedure (11). Typically, this procedure yielded 45–48 % of the lignin from the kraft pulps. The resulting samples were then derivatized and analyzed by NMR. Among the many functional groups that can be analyzed following this protocol, we were able to rapidly determine the amounts of carboxyl and phenolic groups present in the residual lignin. Figure 6 summarizes the changes in carboxylic acid groups from the residual lignin samples isolated from the high-kappa pulps. This analysis indicates that the residual lignin after LMS(E) and LMS(EPO) stages is enriched in acid groups. Differences in carboxyl content for the LMS_{HBT}(EPO) and LMS_{NHAA}(EPO) suggest that the different mediators may be altering the types of chemistry involved in lignin removal.

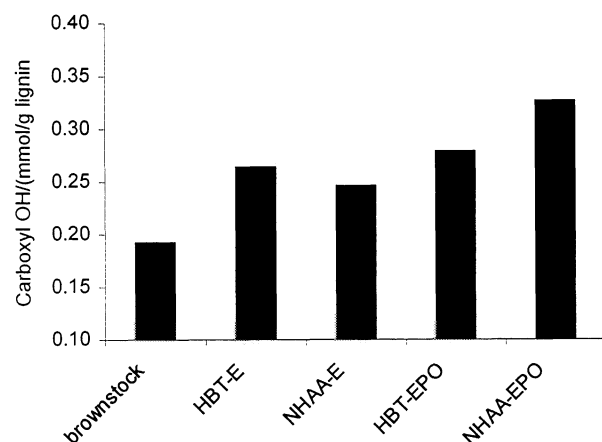


Figure 6. Carboxyl content of the kappa 75.4 SW kraft pulp for brownstock and LMS(E) and LMS(EPO) treated pulps.

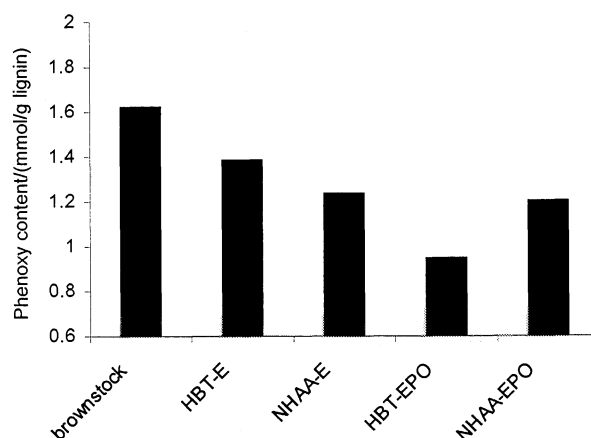


Figure 8. Phenoxyl content of the kappa 75.4 SW kraft pulp for brownstock and LMS(E) and LMS(EPO) treated pulps.

Analysis of the residual lignin from the low kappa brownstock, LMS_{HBT}(EPO) and LMS_{NHAA}(EPO) pulps (see Fig. 7) bears a comparable trend to the high-kappa pulp data. However, the relative differences are amplified for the low-kappa pulps. This result implies that the differences in lignin from the high- and low-kappa pulps affect the delignification chemistry during the LMS(E) stage.

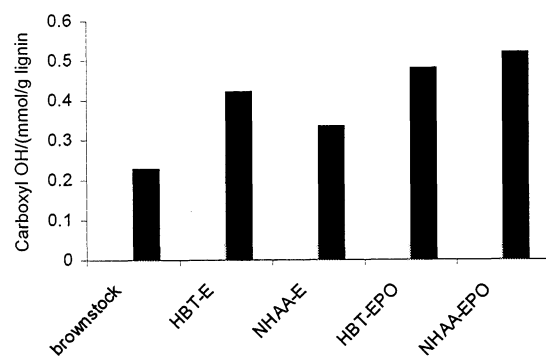


Figure 7. Carboxyl content of the kappa 33.8 SW kraft pulp for brownstock and LMS(E) and LMS(EPO) treated pulps.

The NMR analysis also permitted us to monitor changes in phenoxyl content for the various kraft pulps. The results of this analysis are presented in Figures 8 and 9.

The phenoxyl content analysis of the residual lignins suggests that the LMS(E*) stages consume free phenoxyl groups in lignin. The LMS_{HBT}(EPO) treatment of the high kappa SW kraft pulp removed additional phenoxyl groups, but this depletion was not observed with the LMS_{NHAA}(EPO) sequence.

Interestingly, the loss of phenoxyl groups for the various LMS(E*) stages for the low-kappa SW kraft pulp was more pronounced than the high-kappa pulp. This is an unexpected result since the low kappa pulp was not as bleachable as the high-kappa kraft pulp under the experimental conditions employed in this study.

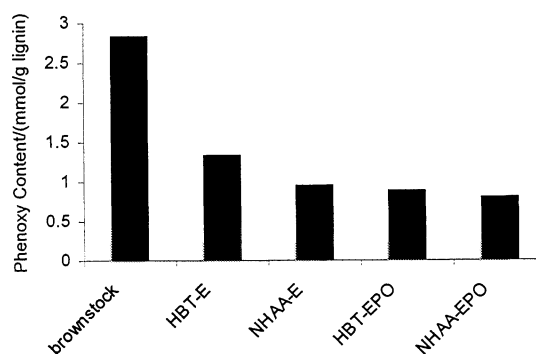


Figure 9. Phenoxyl content of the kappa 33.8 SW kraft pulp for brownstock and LMS(E) and LMS(EPO) treated pulps.

CONCLUSIONS

Our results suggest that the LMS(E*) delignification of kraft pulps can be effectively implemented for high- and low-kappa kraft pulps. The use of additional oxidant in the extraction stage after an LMS-stage is clearly beneficial for both systems. Therefore, this technology could be potentially developed into a viable alternative to oxygen delignification of high-kappa kraft pulps.

The relative changes in carboxyl and phenoxy content for the high- and low-kappa kraft pulps suggest that the nature of residual lignin influences the effectiveness of an LMS(E*) treatment. Furthermore, differences were noted in the structure of residual lignin from LMS_{HBT}(E*) and LMS_{NHAA}(E*). This implies that the different N-hydroxy based mediators may be exhibiting different delignification chemistries. Further studies will be needed to elucidate the complex chemistry of LMS with high-kappa kraft pulps.

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